
**UNO™ Q&S R Continuous
Bed Ion Exchange
Replacement Column
Instruction Manual**

Catalog Numbers

**720-0011, 720-0013, 720-0015,
720-0031, 720-0033 and 720-0035**

BIO-RAD

Introduction

UNO prepacked ion-exchange columns are designed to meet the needs of the bio-chromatographer for rapid and reproducible high-resolution separations of biomolecules including proteins, peptides and polynucleotides. The UNO Replacement columns provide an economical alternative to purchasing a new complete UNO column. The UNO Replacement column hardware utilizes the existing hardware from your worn-out UNO column.

Installing the Replacement Column

Preparing the Used UNO Column (refer to Figure 1)

1. Remove the tube end fittings (2) from the UNO column end caps (3).
2. Remove one end cap (3) from the column and remove glass tube assembly (10) from the clear shield (4).
3. Remove both tan bed supports (5) and spacers (7), if they are still attached, from the glass tube and set aside.
4. Discard the glass tube filled with the used continuous bed support material in accordance to local regulations.

Preparing the New UNO Replacement Column

1. Unthread the end caps (11) and remove UNO Replacement Column glass tube (16) from the clear shield (12).
2. Remove the solid white plug (14) from both ends of the glass tube.

Assembling the New UNO Replacement Column

1. Each of the tan bed supports (5) from the used UNO column should have a 4 mm wide brown spacer (7) located between the o-ring (6) and the top of the bed support. If the bed support does not have the spacer, insert a new spacer, from the UNO Replacement column parts kit, onto the bed support (5).
2. Insert each tan bed support (5) with spacer (7) into each end of the UNO Replacement Column glass tube (6). Push the bed supports into the column until they touch the frits (9).
3. Insert the UNO replacement column glass tube into the clear shield (4) and attach the end caps (3) from the used UNO column onto the shield (4). Tighten the end caps.
4. Attach the tube end fittings (2) to the column and chromatography system and follow equilibration protocol listed in the next section.

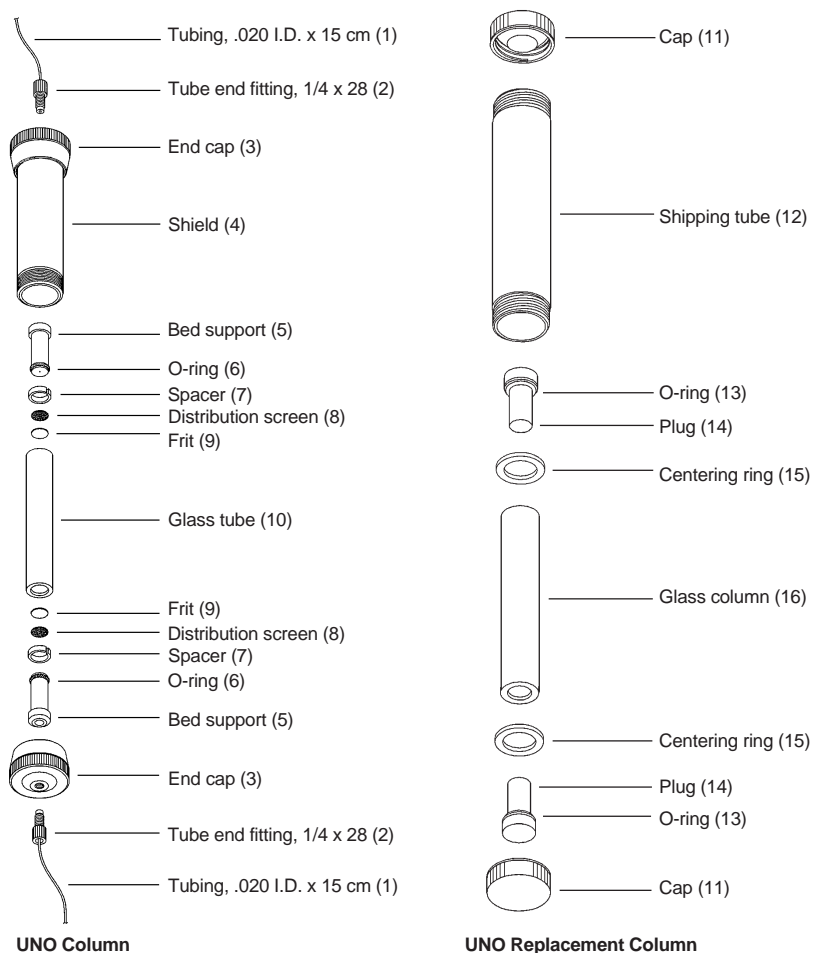


Fig. 1. Drawing of UNO column and UNO replacement column.

The columns are supplied in a storage buffer of 0.1 M NaCl + 20% ethanol. The counter ion for the Q column is Cl⁻ and for the S column is Na⁺. Prior to initial use and after extended storage periods, each column should be conditioned as described below (steps 1–4). Always use HPLC grade reagents and filter and degas the buffers. During this operation do not exceed the following flow rates:

S-1 or Q-1	1 ml/min
S-6 or Q-6	2 ml/min
S-12 or Q-12	3 ml/min

1. Wash with 5 column volumes of water. Elevated backpressures may occur when washing with deionized water. Do not exceed 700 psi.
2. Wash with 5 column volumes of low ionic strength start buffer [*e.g.* 20 mM Tris-HCl (Q) or 20 mM Sodium Phosphate(S)].
3. Wash with 5 column volumes of high ionic strength elution buffer (*e.g.* starting buffer + 1.0 M NaCl).
4. Wash with 5 column volumes of low ionic strength equilibration buffer [*e.g.* 20 mM Tris-HCl (Q) or 20 mM Sodium Phosphate(S)].

The column may now be further equilibrated in start buffer at the desired flow rate.

Table 1. UNO Q Column Characteristics

	Q-1	Q-6	Q-12
Column volume (ml)	1.3	6	12
Recommended max. protein loading (mg)	20	80	160
Recommended flow-rates (ml/min)	0.5 to 5.0	0.5 to 8.0	0.5 to 10.0
Column Dimensions (mm)	7 x 35	12 x 53	15 x 68
Maximum operating pressure (psi/MPa/bar)	700/4.5/48	700/4.5/48	700/4.5/48

Table 2. UNO S Column Characteristics

	S-1	S-6	S-12
Column volume (ml)	1.3	6	12
Recommended max. protein loading (mg)	20	80	160
Recommended flow-rates (ml/min)	0.5 to 6.0	0.5 to 8.0	0.5 to 10.0
Column Dimensions (mm)	7 x 35	12 x 53	15 x 68
Maximum operating pressure (psi/MPa/bar)	700/4.5/48	700/4.5/48	700/4.5/48

Column Hygiene

Careful preparation (especially filtration) of the sample and the buffers will maintain the column performance and lifetime. Normally, washing with 1.0 M NaCl or KCl will remove most bound components. However, if there is a significant decrease in column performance (*i.e.* increasing backpressures or a significant drop in resolution) then a more extensive cleaning protocol as described below (steps 1–6) should be used. Always reverse the flow during this procedure so tightly-bound substances at the top of the column are quickly removed.

This process should be performed within the following flow rates.

S-1 or Q-1	1 ml/min
S-6 or Q-6	2 ml/min
S-12 or Q-12	3 ml/min

1. Wash with 2 column volumes of 2.0 M NaCl or KCl.
2. Wash with 1 column volume of 2.0 M NaOH followed by 3 column volumes of 0.1 M NaCl.
3. Wash with 1 column volume of 50% acetic acid followed by 3 column volumes of 0.1 M NaCl. Wash with 1 column volume of deionized water.
4. If lipid contamination is a problem, wash with 1 column volume of MeOH followed by 1 column volume of deionized water and 3 column volumes of 0.1 M NaCl.
5. Wash with 2 column volumes of 2.0 M NaCl or KCl or the salt containing the desired counter-ion.
6. Change the top frit and wash with 3 column volumes of start buffer.

References

1. Kopaciewicz, W., Rounds, M. A., Fausnaugh, J. and Regnier, F. E. Retention Model for High-performance Ion-Exchange Chromatography, *J. Chromatography*, **266**, 3-21 (1983).
2. A Guide to the Properties and Uses of Detergents in Biology and Biochemistry. J. Neugebauer (Calbiochem Biochemicals).

Product Information

Catalog Number	Product Description
720-0001	UNO Q-1 Column
720-0003	UNO Q-6 Column
720-0005	UNO Q-12 Column
720-0009	UNO Q Polishing Column
720-0011	UNO Q-1 R Column , replacement column
720-0013	UNO Q-6 R Column , replacement column
720-0015	UNO Q-12 R Column , replacement column
720-0021	UNO S-1 Column
720-0023	UNO S-6 Column
720-0025	UNO S-12 Column
720-0029	UNO S Polishing Column
720-0031	UNO S-1 R Column , replacement column
720-0033	UNO S-6 R Column , replacement column
720-0035	UNO S-12 R Column , replacement column
751-0091	Bio-Scale 2 Replacement Part Kit , includes 5 frits, 5 distribution screens, 2 o-rings, 1 frit remover. Use this kit for the UNO S-1 or Q-1 Column.
751-0095	Bio-Scale 10 Replacement Part Kit , includes 5 frits, 5 distribution screens, 2 o-rings, 1 frit remover. Use this kit for the UNO S-6 or Q-6 Column.
751-0097	Bio-Scale 20 Replacement Part Kit , includes 5 frits, 5 distribution screens, 2 o-rings, 1 frit remover. Use this kit for the UNO S-12 or Q-12 Column.
751-0099	Bio-Scale Fittings Kit , includes 2 Super Flangeless Nuts (1/4 x 28 threads) and 6 ferrules, 2 Flangeless M6 Nuts, 4 ferrules and 2 caps, 2 Fingertight II fittings (10-32 threads). Use this kit for any UNO Column.
125-0561	Anion Exchange Standards , 6 pack
125-0562	Cation Exchange Standards , 6 pack

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